Huanglongbing: new epidemic situation in the French West Indies, caused by Candidatus *Liberibacter asiaticus*

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alimentation, environnement, travail

Presentation

Huanglongbing is an unculturable vascular citrus bacterium transmitted from infected to healthy plants through grafting or by citrus psyllids *Diaphorina citri* and *Trioza erytreae*. Three species are associated based on the 16S rDNA sequence analysis:

- Candidatus *Liberibacter asiaticus* (Las)
- Ca. *L. africanus* (Laf)
- Ca. *L. americanus* (Lam)

Lobsided Tahiti lime with curved columella
(Photo by Caroline Sylvaniélo)

Blotchy mottle pattern symptoms
(Photos by Gilles Cellier)

Severe leaf and fruit drop
(Photos by Gilles Cellier)

The Asian citrus psyllid, *D. citri*, is a sap-sucking, hemipteran bug.
It is one of only two confirmed vectors of Huanglongbing, widely distributed in southern Asia and has spread to other citrus growing regions: Middle East, America, and the Caribbean.
Currently absent in Europe.

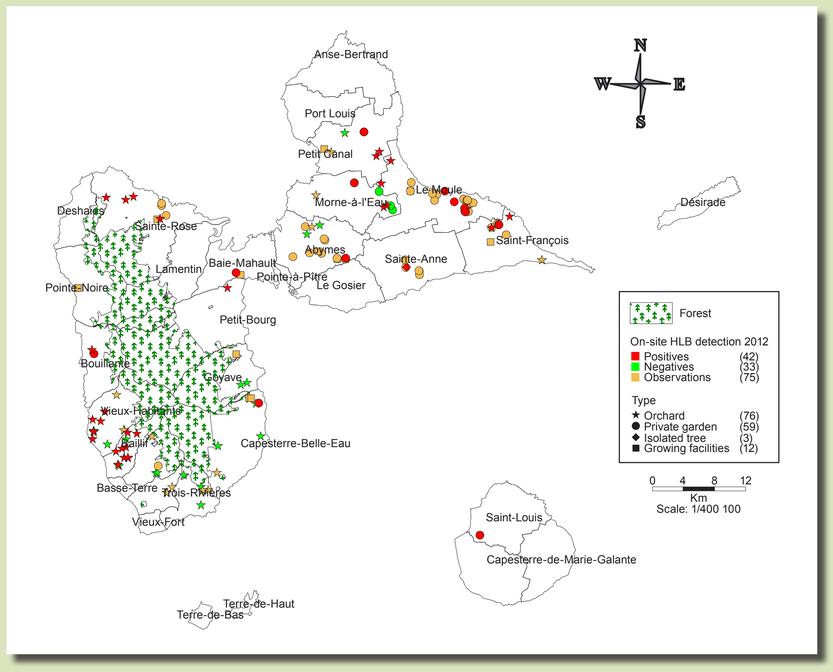
Aduld psyllid *Diaphorina citri* along with nymph producing waxy exudate
(Photo by Michael E. Rogers)

Methodology

Since 2012, a total of 450 sites were prospected in Martinique and Guadeloupe, where 20 leaves from 10-30 trees were analyzed. DNA extraction was performed (DNeasy Plant Mini Kit, Qiagen) on fresh or dried leaf midribs, along with negative control midribs (Citrus paradisi “Star Rubis”) and polymerase chain reaction amplification was done with the species-specific primers, A2/J5 (Hocquellet et al. 1999) and GB1/GB3 (Teixeira et al. 2005).

Only Las-specific 703 bp amplicons were obtained and 20 (out of 43) were sequenced (Beckman Coulter Genomics, United Kingdom; Genbank accession : KF699074 to KF699093) and blasted against NCBI-nr.

Guadeloupe



Guadeloupe: HLB Situation in 2012 (*source: DAAF 971)*

Las was detected in Guadeloupe on March 2012 at Le Moule (East) in a Tahiti lime orchard (*C. latifolia*) and crossed the island in six months.

D. citri was detected in 1998, where the control of the the psyllid population has been effective with *Tamarixia radiata*.

Results

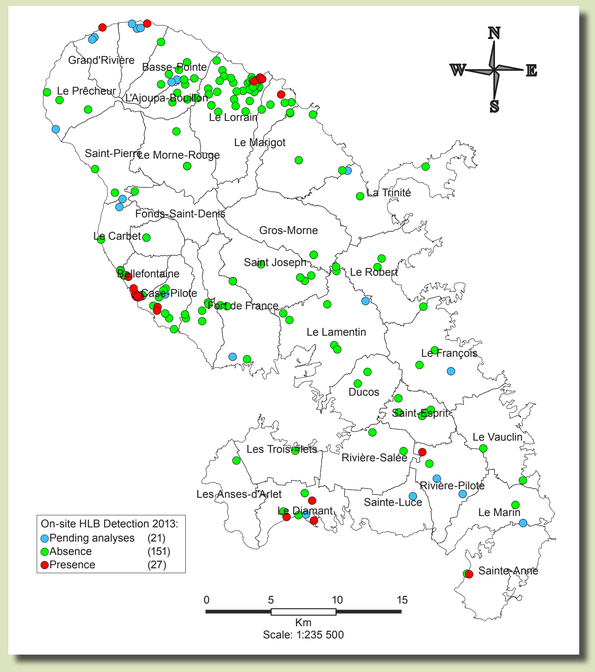
Based on MOA033

Detection and caracterisation of Candidatus *Liberibacter asiaticus*.

BLAST analysis revealed 100% identity with the 50S ribosomal protein subunit L1 (rplA) and L10 (rplJ) and no significant homology to other organisms.

Sequence assembly on a reference genome (NC_012985) showed 100% homology.

Martinique



Martinique: HLB Situation in 2013 (*source: DAAF 972)*

Las was detected in Martinique on May 2013 on Tahiti lime (*C. latifolia*) at Bellefontaine (NW / private garden) and at Le Lorrain (NE / orchard).

D. citri was detected in 2012 and presents a mean parasitism rate of 70% by *Tamarixia radiata*.

Distribution map of Candidatus *Liberibacter asiaticus*
(source: EPPO)

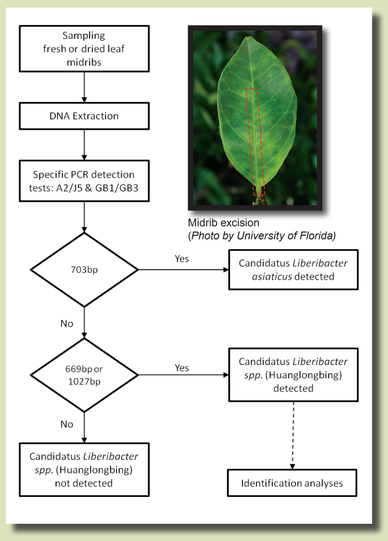
Method Assessment

Anses-LSV unit for tropical pest is the National Reference Laboratory on tropical phytopathogenic bacteria and viruses. It has produced and validated HLB detection methods (MOA033), under ISO 17025 accreditation.

Based on Li & al. duplex PCR (A2/J5 & GB1/GB3) was assessed on several criteria to detect Las, but also Lam & Laf:

- Sensitivity
- Specificity
- Accuracy
- Repetability
- Detectability
- Processed speed
- Transferability
- Final score: 6.47 / 7

HLB detection scheme from MOA033



Midrib excision
(Photo by University of Florida)

Conclusion

- First report of Candidatus *Liberibacter asiaticus* in the French West Indies
- Disease management needs to especially target the vector psyllid
- Synergy between local NPPO & LSV-RAPT Reference laboratory

This report raises the importance of plant certification, psyllid population control, and surveillance of territories close to the French West Indies, with regards to the risk that HLB presents to citrus production worldwide.

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